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(54) Title: NUCLEOSIDE DERIVATIVES AS INHIBITORS OF RNA-DEPENDENT RNA VIRAL POLYMERASE

(57) **Abstract:** The present invention provides nucleoside compounds and certain derivatives thereof which are inhibitors of RNA-dependent RNA viral polymerase. These compounds are inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection. They are particularly useful as inhibitors of hepatitis C virus (HCV) NS5B polymerase, as inhibitors of HCV replication, and/or for the treatment of hepatitis C infection. The invention also describes pharmaceutical compositions containing such nucleoside compounds alone or in combination with other agents active against RNA-dependent RNA viral infection, in particular HCV infection. Also disclosed are methods of inhibiting RNA-dependent RNA polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection with the nucleoside compounds of the present invention.

NUCLEOSIDE DERIVATIVES AS INHIBITORS OF RNA-DEPENDENT RNA VIRAL POLYMERASE

FIELD OF THE INVENTION

The present invention is concerned with nucleoside compounds and certain derivatives thereof, their synthesis, and their use as inhibitors of RNA-dependent RNA viral polymerase. The compounds of the present invention are inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection. They are particularly useful as inhibitors of hepatitis C virus (HCV) NS5B polymerase, as inhibitors of HCV replication, and for the treatment of hepatitis C infection.

BACKGROUND OF THE INVENTION

Hepatitis C virus (HCV) infection is a major health problem that leads to chronic liver disease, such as cirrhosis and hepatocellular carcinoma, in a substantial number of infected individuals, estimated to be 2-15% of the world's population. There are an estimated 4.5 million infected people in the United States alone, according to the U.S. Center for Disease Control. According to the World Health Organization, there are more than 200 million infected individuals worldwide, with at least 3 to 4 million people being infected each year. Once infected, about 20% of people clear the virus, but the rest harbor HCV the rest of their lives. Ten to twenty percent of chronically infected individuals eventually develop liver-destroying cirrhosis or cancer. The viral disease is transmitted parenterally by contaminated blood and blood products, contaminated needles, or sexually and vertically from infected mothers or carrier mothers to their off-spring. Current treatments for HCV infection, which are restricted to immunotherapy with recombinant interferon- α alone or in combination with the nucleoside analog ribavirin, are of limited clinical benefit. Moreover, there is no established vaccine for HCV. Consequently, there is an urgent need for improved therapeutic agents that effectively combat chronic HCV infection. The state of the art in the treatment of HCV infection has been reviewed, and reference is made to the following publications: B. Dymock, et al., "Novel approaches to the treatment of hepatitis C virus infection," Antiviral Chemistry & Chemotherapy, 11: 79-96 (2000); H. Rosen, et al., "Hepatitis C virus: current understanding and prospects for future therapies," Molecular Medicine Today, 5: 393-

399 (1999); D. Moradpour, et al., "Current and evolving therapies for hepatitis C," European J. Gastroenterol. Hepatol., 11: 1189-1202 (1999); R. Bartenschlager, "Candidate Targets for Hepatitis C Virus-Specific Antiviral Therapy," Intervirology, 40: 378-393 (1997); G.M. Lauer and B.D. Walker, "Hepatitis C Virus Infection," N. Engl. J. Med., 345: 41-52 (2001); B.W. Dymock, "Emerging therapies for hepatitis C virus infection," Emerging Drugs, 6: 13-42 (2001); and C. Crabb, "Hard-Won Advances Spark Excitement about Hepatitis C," Science: 506-507 (2001); the contents of all of which are incorporated by reference herein in their entirety.

Different approaches to HCV therapy have been taken, which include
10 the inhibition of viral serine proteinase (NS3 protease), helicase, and RNA-dependent RNA polymerase (NS5B), and the development of a vaccine.

The HCV virion is an enveloped positive-strand RNA virus with a single oligoribonucleotide genomic sequence of about 9600 bases which encodes a polyprotein of about 3,010 amino acids. The protein products of the HCV gene
15 consist of the structural proteins C, E1, and E2, and the non-structural proteins NS2, NS3, NS4A and NS4B, and NS5A and NS5B. The nonstructural (NS) proteins are believed to provide the catalytic machinery for viral replication. The NS3 protease releases NS5B, the RNA-dependent RNA polymerase from the polyprotein chain. HCV NS5B polymerase is required for the synthesis of a double-stranded RNA from
20 a single-stranded viral RNA that serves as a template in the replication cycle of HCV. NS5B polymerase is therefore considered to be an essential component in the HCV replication complex [see K. Ishi, et al., "Expression of Hepatitis C Virus NS5B Protein: Characterization of Its RNA Polymerase Activity and RNA Binding," Hepatology, 29: 1227-1235 (1999) and V. Lohmann, et al., "Biochemical and Kinetic
25 Analyses of NS5B RNA-Dependent RNA Polymerase of the Hepatitis C Virus," Virology, 249: 108-118 (1998)]. Inhibition of HCV NS5B polymerase prevents formation of the double-stranded HCV RNA and therefore constitutes an attractive approach to the development of HCV-specific antiviral therapies.

It has now been found that nucleoside compounds of the present
30 invention and certain derivatives thereof are potent inhibitors of RNA-dependent RNA viral replication and in particular HCV replication. The 5'-triphosphate derivatives of these nucleoside compounds are inhibitors of RNA-dependent RNA viral polymerase and in particular HCV NS5B polymerase. The instant nucleoside compounds and derivatives thereof are useful to treat RNA-dependent RNA viral
35 infection and in particular HCV infection.

It is therefore an object of the present invention to provide nucleoside compounds and certain derivatives thereof which are useful as inhibitors of RNA-dependent RNA viral polymerase and in particular as inhibitors of HCV NS5B polymerase.

5 It is another object of the present invention to provide nucleoside compounds and certain derivatives thereof which are useful as inhibitors of the replication of an RNA-dependent RNA virus and in particular as inhibitors of the replication of hepatitis C virus.

10 It is another object of the present invention to provide nucleoside compounds and certain derivatives thereof which are useful in the treatment of RNA-dependent RNA viral infection and in particular in the treatment of HCV infection.

It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds of the present invention in association with a pharmaceutically acceptable carrier.

15 It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof of the present invention for use as inhibitors of RNA-dependent RNA viral polymerase and in particular as inhibitors of HCV NS5B polymerase.

20 It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof of the present invention for use as inhibitors of RNA-dependent RNA viral replication and in particular as inhibitors of HCV replication.

25 It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof of the present invention for use in the treatment of RNA-dependent RNA viral infection and in particular in the treatment of HCV infection.

30 It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof of the present invention in combination with other agents active against an RNA-dependent RNA virus and in particular against HCV.

It is another object of the present invention to provide methods for the inhibition of RNA-dependent RNA viral polymerase and in particular for the inhibition of HCV NS5B polymerase.

It is another object of the present invention to provide methods for the inhibition of RNA-dependent RNA viral replication and in particular for the inhibition of HCV replication.

It is another object of the present invention to provide methods for the treatment of RNA-dependent RNA viral infection and in particular for the treatment of HCV infection.

It is another object of the present invention to provide methods for the treatment of RNA-dependent RNA viral infection in combination with other agents active against RNA-dependent RNA virus and in particular for the treatment of HCV infection in combination with other agents active against HCV.

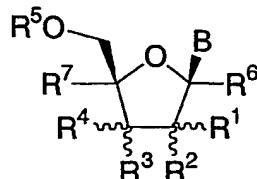
It is another object of the present invention to provide nucleoside compounds and certain derivatives thereof and their pharmaceutical compositions for use as a medicament for the inhibition of RNA-dependent RNA viral replication and/or the treatment of RNA-dependent RNA viral infection and in particular for the inhibition of HCV replication and/or the treatment of HCV infection.

It is another object of the present invention to provide for the use of the nucleoside compounds and certain derivatives thereof of the present invention and their pharmaceutical compositions for the manufacture of a medicament for the inhibition of RNA-dependent RNA viral replication and/or the treatment of RNA-dependent RNA viral infection and in particular for the inhibition of HCV replication and/or the treatment of HCV infection.

These and other objects will become readily apparent from the detailed description which follows.

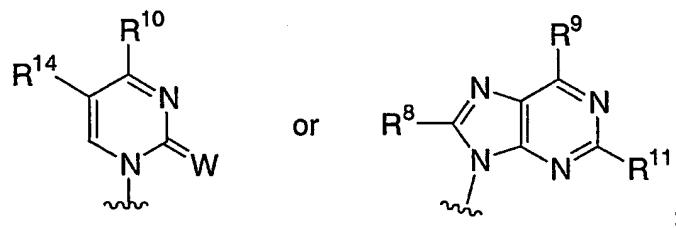
25 SUMMARY OF THE INVENTION

The present invention relates to compounds of structural formula I of the indicated stereochemical configuration:



(1)

or a pharmaceutically acceptable salt thereof;
wherein B is



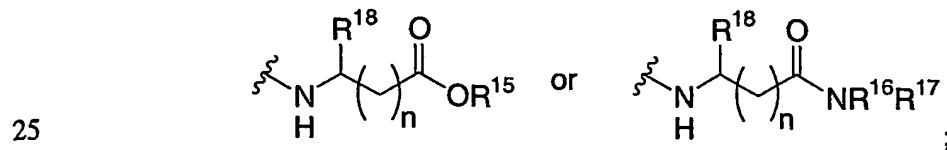
5 W is O or S;
R¹ is fluoromethyl, difluoromethyl, or trifluoromethyl;
R² is hydrogen, fluorine, amino, hydroxy, mercapto, C₁-4 alkoxy, C₁-8 alkylcarbonyloxy, or C₁-4 alkyl;

10 R³ and R⁴ are each independently selected from the group consisting of hydrogen, cyano, azido, halogen, hydroxy, mercapto, amino, C₁-4 alkoxy, C₁-8 alkylcarbonyloxy, C₂-4 alkenyl, C₂-4 alkynyl, and C₁-4 alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C₁-4 alkoxy, C₁-4 alkylthio, or one to three fluorine atoms;

15 R⁵ is hydrogen, C₁-10 alkylcarbonyl, P₃O₉H₄, P₂O₆H₃, or P(O)R₁₂R₁₃;

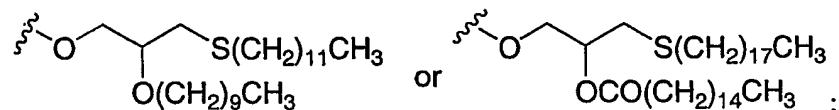
R⁶ and R⁷ are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl;
R⁸ is hydrogen, C₁-4 alkyl, C₂-4 alkynyl, halogen, cyano, carboxy, C₁-4 alkylcarbonyl, azido, amino, C₁-4 alkylamino, di(C₁-4 alkyl)amino, hydroxy, C₁-6 alkoxy, C₁-6 alkylthio, C₁-6 alkylsulfonyl, or (C₁-4 alkyl)O-2 aminomethyl;

20 R⁹ and R¹⁰ are each independently hydrogen, hydroxy, mercapto, halogen, C₁-4 alkoxy, C₁-4 alkylthio, C₁-8 alkylcarbonyloxy, C₃-6 cycloalkylcarbonyloxy, C₁-8 alkylcarbonyloxy, C₃-6 cycloalkyloxycarbonyloxy, OCH₂CH₂SC(=O)C₁-4 alkyl, OCH₂O(C=O)C₁-4 alkyl, OCH(C₁-4 alkyl)O(C=O)C₁-4 alkyl, amino, C₁-4 alkylamino, di(C₁-4 alkyl)amino, C₃-6 cycloalkylamino, di(C₃-6 cycloalkyl)amino, or an amino acyl residue having structural formula



n is 0, 1, or 2;

R₁₁ is hydrogen, hydroxy, halogen, C₁₋₄ alkoxy, amino, C₁₋₄ alkylamino, di(C₁₋₄ alkyl)amino, C₃₋₆ cycloalkylamino, or di(C₃₋₆ cycloalkylamino);
 R₁₅, R₁₆, and R₁₇ are each independently hydrogen or C₁₋₆ alkyl;
 R₁₂ and R₁₃ are each independently hydroxy, -OCH₂CH₂SC(=O)C₁₋₄ alkyl,
 5 -OCH₂O(C=O)OC₁₋₄ alkyl, -NHCHMeCO₂Me, -OCH(C₁₋₄ alkyl)O(C=O)C₁₋₄ alkyl,



R₁₄ is hydrogen, C₁₋₆ alkyl, C₂₋₆ alkenyl, C₂₋₆ alkynyl, C₁₋₄ alkylamino, CF₃, or halogen; and
 10 R₁₈ is hydrogen, C₁₋₄ alkyl, or phenyl C₀₋₂ alkyl.

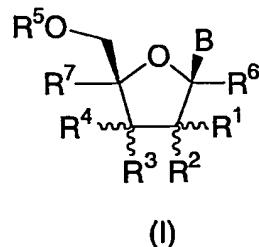
The compounds of formula I are useful as inhibitors of RNA-dependent RNA viral polymerase and in particular of HCV NS5B polymerase. They are also inhibitors of RNA-dependent RNA viral replication and in particular of HCV replication and are useful for the treatment of RNA-dependent RNA viral infection
 15 and in particular for the treatment of HCV infection.

Also encompassed within the present invention are pharmaceutical compositions containing the compounds alone or in combination with other agents active against RNA-dependent RNA virus and in particular against HCV as well as methods for the inhibition of RNA-dependent RNA viral replication and for the
 20 treatment of RNA-dependent RNA viral infection.

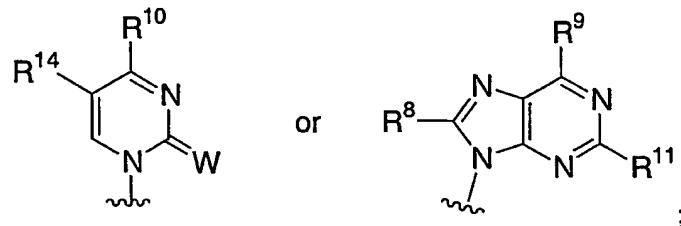
DETAILED DESCRIPTION OF THE INVENTION

The present invention relates to compounds of structural formula I of the indicated stereochemical configuration:

25



or a pharmaceutically acceptable salt thereof;
wherein B is

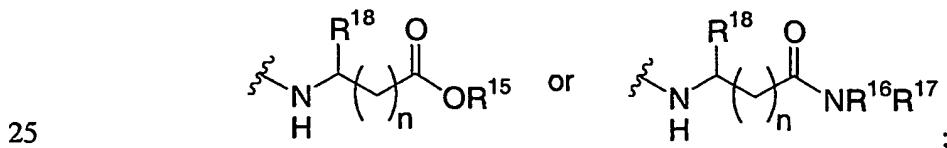


5 W is O or S;
R1 is fluoromethyl, difluoromethyl, or trifluoromethyl;
R2 is hydrogen, fluorine, amino, hydroxy, mercapto, C1-4 alkoxy, C1-8 alkylcarbonyloxy, or C1-4 alkyl;

10 R3 and R4 are each independently selected from the group consisting of hydrogen, cyano, azido, halogen, hydroxy, mercapto, amino, C1-4 alkoxy, C1-8 alkylcarbonyloxy, C2-4 alkenyl, C2-4 alkynyl, and C1-4 alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C1-4 alkoxy, C1-4 alkylthio, or one to three fluorine atoms;

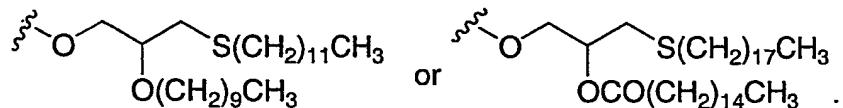
15 R5 is hydrogen, C1-10 alkylcarbonyl, P3O9H4, P2O6H3, or P(O)R12R13;

20 R6 and R7 are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; R8 is hydrogen, C1-4 alkyl, C2-4 alkynyl, halogen, cyano, carboxy, C1-4 alkyloxycarbonyl, azido, amino, C1-4 alkylamino, di(C1-4 alkyl)amino, hydroxy, C1-6 alkoxy, C1-6 alkylthio, C1-6 alkylsulfonyl, or (C1-4 alkyl)0-2 aminomethyl; R9 and R10 are each independently hydrogen, hydroxy, mercapto, halogen, C1-4 alkoxy, C1-4 alkylthio, C1-8 alkylcarbonyloxy, C3-6 cycloalkylcarbonyloxy, C1-8 alkyloxycarbonyloxy, C3-6 cycloalkyloxycarbonyloxy, -OCH2CH2SC(=O)C1-4 alkyl, -OCH2O(C=O)C1-4 alkyl, -OCH(C1-4 alkyl)O(C=O)C1-4 alkyl, amino, C1-4 alkylamino, di(C1-4 alkyl)amino, C3-6 cycloalkylamino, di(C3-6 cycloalkyl)amino, or an amino acyl residue having structural formula



n is 0, 1, or 2;

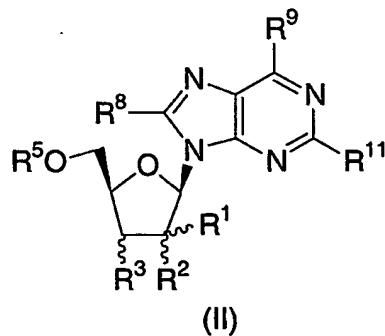
R¹¹ is hydrogen, hydroxy, halogen, C₁₋₄ alkoxy, amino, C₁₋₄ alkylamino, di(C₁₋₄ alkyl)amino, C₃₋₆ cycloalkylamino, or di(C₃₋₆ cycloalkylamino);
 R¹⁵, R¹⁶, and R¹⁷ are each independently hydrogen or C₁₋₆ alkyl;
 R¹² and R¹³ are each independently hydroxy, -OCH₂CH₂SC(=O)C₁₋₄ alkyl,
 5 -OCH₂O(C=O)OC₁₋₄ alkyl, -NHCHMeCO₂Me, -OCH(C₁₋₄ alkyl)O(C=O)C₁₋₄ alkyl,



R¹⁴ is hydrogen, C₁₋₆ alkyl, C₂₋₆ alkenyl, C₂₋₆ alkynyl, C₁₋₄ alkylamino, CF₃, or halogen; and
 10 R¹⁸ is hydrogen, C₁₋₄ alkyl, or phenyl C₀₋₂ alkyl.

The compounds of formula I are useful as inhibitors of RNA-dependent RNA viral polymerase. They are also inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection.

15 In one embodiment of the compounds of structural formula I are the compounds of structural formula II:



or a pharmaceutically acceptable salt thereof;
 wherein
 20 R¹ is fluoromethyl or difluoromethyl;
 R² is hydroxy, fluoro, or C₁₋₃ alkoxy;
 R³ is hydrogen, halogen, hydroxy, amino, or C₁₋₃ alkoxy;
 R⁵ is hydrogen, P₃O₉H₄, P₂O₆H₃, or PO₃H₂;

R⁸ is hydrogen, amino, or C₁-4 alkylamino; and

R⁹ and R¹¹ are each independently hydrogen, halogen, hydroxy, amino, C₁-4 alkylamino, di(C₁-4 alkyl)amino, or C₃-6 cycloalkylamino.

In a second embodiment of the compounds of structural formula I are

5 the compounds of structural formula II wherein:

R¹ is fluoromethyl or difluoromethyl;

R² is hydroxy, fluoro, or methoxy;

R³ is hydrogen, fluoro, hydroxy, amino, or methoxy;

R⁵ is hydrogen or P₃O₉H₄;

10 R⁸ is hydrogen or amino; and

R⁹ and R¹¹ are each independently hydrogen, fluoro, hydroxy, or amino.

Illustrative, but nonlimiting, examples of compounds of the present invention of structural formula I which are useful as inhibitors of RNA-dependent RNA viral polymerase are the following:

15 6-amino-9-(2-C-fluoromethyl- β -D-ribofuranosyl)purine;

6-amino-9-(2-C-fluoromethyl- β -D-arabinofuranosyl)purine;

2-amino-9-(2-C-fluoromethyl- β -D-ribofuranosyl)-3,9-dihydropurin-6-one;

2-amino-9-(2-C-fluoromethyl- β -D-arabinofuranosyl)-3,9-dihydropurin-6-one;

2-amino-9-(2-C-fluoromethyl- β -D-ribofuranosyl)-3,9-dihydropurin-6-thione;

20 2,6-diamino-9-(2-C-fluoromethyl- β -D-ribofuranosyl)purine;

9-(2-C-fluoromethyl- β -D-ribofuranosyl)-6-methylaminopurine;

2'-C-(fluoromethyl)cytidine;

2'-C-(fluoromethyl)-5-methylcytidine;

2'-C-(fluoromethyl)uridine; and

25 2'-C-(fluoromethyl)-5-methyluridine;

and the corresponding 5'-triphosphates;

or a pharmaceutically acceptable salt thereof.

In one embodiment of the present invention, the nucleoside compounds of the present invention are useful as inhibitors of positive-sense single-stranded

30 RNA-dependent RNA viral polymerase, inhibitors of positive-sense single-stranded RNA-dependent RNA viral replication, and/or for the treatment of positive-sense single-stranded RNA-dependent RNA viral infection. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA virus is a *Flaviviridae* virus or a *Picornaviridae* virus. In a subclass of this class, the *Picornaviridae* virus is a

rhinovirus, a poliovirus, or a hepatitis A virus. In a second subclass of this class, the *Flaviviridae* virus is selected from the group consisting of hepatitis C virus, yellow fever virus, dengue virus, West Nile virus, Japanese encephalitis virus, Banzi virus, and bovine viral diarrhea virus (BVDV). In a subclass of this subclass, the

5 *Flaviviridae* virus is hepatitis C virus.

Another aspect of the present invention is concerned with a method for inhibiting RNA-dependent RNA viral polymerase, a method for inhibiting RNA-dependent RNA viral replication, and/or a method for treating RNA-dependent RNA viral infection in a mammal in need thereof comprising administering to the mammal
10 a therapeutically effective amount of a compound of structural formula I.

In one embodiment of this aspect of the present invention, the RNA-dependent RNA viral polymerase is a positive-sense single-stranded RNA-dependent RNA viral polymerase. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA viral polymerase is a *Flaviviridae* viral polymerase or
15 a *Picornaviridae* viral polymerase. In a subclass of this class, the *Picornaviridae* viral polymerase is rhinovirus polymerase, poliovirus polymerase, or hepatitis A virus polymerase. In a second subclass of this class, the *Flaviviridae* viral polymerase is selected from the group consisting of hepatitis C virus polymerase, yellow fever virus polymerase, dengue virus polymerase, West Nile virus polymerase, Japanese
20 encephalitis virus polymerase, Banzi virus polymerase, and bovine viral diarrhea virus (BVDV) polymerase. In a subclass of this subclass, the *Flaviviridae* viral polymerase is hepatitis C virus polymerase.

In a second embodiment of this aspect of the present invention, the RNA-dependent RNA viral replication is a positive-sense single-stranded RNA-dependent RNA viral replication. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA viral replication is *Flaviviridae* viral replication or *Picornaviridae* viral replication. In a subclass of this class, the *Picornaviridae* viral replication is rhinovirus replication, poliovirus replication, or hepatitis A virus replication. In a second subclass of this class, the *Flaviviridae* viral replication is
25 selected from the group consisting of hepatitis C virus replication, yellow fever virus replication, dengue virus replication, West Nile virus replication, Japanese encephalitis virus replication, Banzi virus replication, and bovine viral diarrhea virus replication. In a subclass of this subclass, the *Flaviviridae* viral replication is hepatitis C virus replication.

In a third embodiment of this aspect of the present invention, the RNA-dependent RNA viral infection is a positive-sense single-stranded RNA-dependent viral infection. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA viral infection is *Flaviviridae* viral infection or *Picornaviridae* viral infection. In a subclass of this class, the *Picornaviridae* viral infection is rhinovirus infection, poliovirus infection, or hepatitis A virus infection. In a second subclass of this class, the *Flaviviridae* viral infection is selected from the group consisting of hepatitis C virus infection, yellow fever virus infection, dengue virus infection, West Nile virus infection, Japanese encephalitis virus infection, Banzi virus infection, and bovine viral diarrhea virus infection. In a subclass of this subclass, the *Flaviviridae* viral infection is hepatitis C virus infection.

Throughout the instant application, the following terms have the indicated meanings:

The alkyl groups specified above are intended to include those alkyl groups of the designated length in either a straight or branched configuration. Exemplary of such alkyl groups are methyl, ethyl, propyl, isopropyl, butyl, sec-butyl, tertiary butyl, pentyl, isopentyl, hexyl, isohexyl, and the like.

The term "alkenyl" shall mean straight or branched chain alkenes of two to six total carbon atoms, or any number within this range (e.g., ethenyl, propenyl, butenyl, pentenyl, etc.).

The term "alkynyl" shall mean straight or branched chain alkynes of two to six total carbon atoms, or any number within this range (e.g., ethynyl, propynyl, butynyl, pentynyl, etc.).

The term "cycloalkyl" shall mean cyclic rings of alkanes of three to eight total carbon atoms, or any number within this range (i.e., cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, or cyclooctyl).

The term "cycloheteroalkyl" is intended to include non-aromatic heterocycles containing one or two heteroatoms selected from nitrogen, oxygen and sulfur. Examples of 4-6-membered cycloheteroalkyl include azetidinyl, pyrrolidinyl, piperidinyl, morpholinyl, thiamorpholinyl, imidazolidinyl, tetrahydrofuranyl, tetrahydropyranyl, tetrahydrothiophenyl, piperazinyl, and the like.

The term "alkoxy" refers to straight or branched chain alkoxides of the number of carbon atoms specified (e.g., C₁₋₄ alkoxy), or any number within this range [i.e., methoxy (MeO-), ethoxy, isopropoxy, etc.].

The term "alkylthio" refers to straight or branched chain alkylsulfides of the number of carbon atoms specified (e.g., C₁₋₄ alkylthio), or any number within this range [i.e., methylthio (MeS-), ethylthio, isopropylthio, etc.].

5 The term "alkylamino" refers to straight or branched alkylamines of the number of carbon atoms specified (e.g., C₁₋₄ alkylamino), or any number within this range [i.e., methylamino, ethylamino, isopropylamino, t-butylamino, etc.].

The term "cycloalkylamino" refers to saturated aminohydrocarbons containing one ring of the number of carbon atoms specified (e.g., C₃₋₆ cycloalkylamino), or any number within this range [i.e., cyclopropylamino, 10 cyclobutylamino, cyclopentylamino, and cyclohexylamino].

The term "alkylsulfonyl" refers to straight or branched chain alkylsulfones of the number of carbon atoms specified (e.g., C₁₋₆ alkylsulfonyl), or any number within this range [i.e., methylsulfonyl (MeSO₂-), ethylsulfonyl, isopropylsulfonyl, etc.].

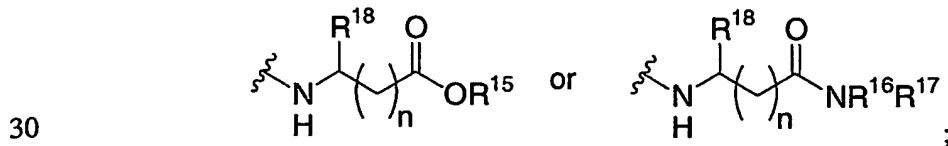
15 The term "alkyloxycarbonyl" refers to straight or branched chain esters of a carboxylic acid derivative of the present invention of the number of carbon atoms specified (e.g., C₁₋₄ alkyloxycarbonyl), or any number within this range [i.e., methyloxycarbonyl (MeOCO-), ethyloxycarbonyl, or butyloxycarbonyl].

20 The term "aryl" includes both phenyl, naphthyl, and pyridyl. The aryl group is optionally substituted with one to three groups independently selected from C₁₋₄ alkyl, halogen, cyano, nitro, trifluoromethyl, C₁₋₄ alkoxy, and C₁₋₄ alkylthio.

The term "halogen" is intended to include the halogen atoms fluorine, chlorine, bromine and iodine.

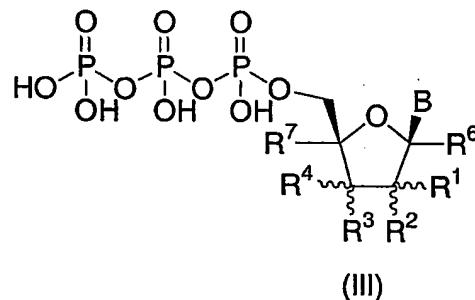
25 The term "substituted" shall be deemed to include multiple degrees of substitution by a named substituent. Where multiple substituent moieties are disclosed or claimed, the substituted compound can be independently substituted by one or more of the disclosed or claimed substituent moieties, singly or plurally.

The term "amino acyl residue" refers to an α -, β -, or γ -amino acyl group of structural formula

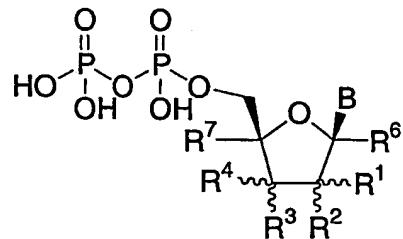
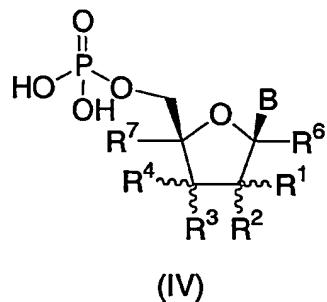


wherein n is 0, 1, or 2 and R₁₅, R₁₆, R₁₇, and R₁₈ are as defined hereinabove. When R₁₈ is not hydrogen, the amino acyl residue contains an asymmetric center and is intended to include the individual *R*- and *S*-enantiomers as well as *RS*-racemic mixtures.

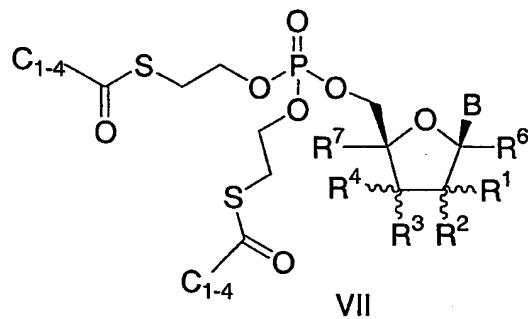
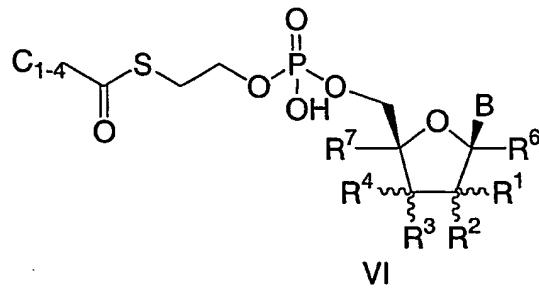
5 The term "5'-triphosphate" refers to a triphosphoric acid ester derivative of the 5'-hydroxyl group of a nucleoside compound of the present invention having the following general structural formula III:



10 wherein B and R₁-R₁₁ are as defined above. The compounds of the present invention are also intended to include pharmaceutically acceptable salts of the triphosphate ester as well as pharmaceutically acceptable salts of 5'-monophosphate and 5'-diphosphate ester derivatives of the structural formulae IV and V, respectively,



15 The term "5'-(S-acyl-2-thioethyl)phosphate" or "SATE" refers to a mono- or di-ester derivative of a 5'-monophosphate nucleoside derivative of the present invention of structural formulae VI and VII, respectively, as well as pharmaceutically acceptable salts of the mono-ester,



The term "composition", as in "pharmaceutical composition," is intended to encompass a product comprising the active ingredient(s) and the inert ingredient(s) that make up the carrier, as well as any product which results, directly or 5 indirectly, from combination, complexation or aggregation of any two or more of the ingredients, or from dissociation of one or more of the ingredients, or from other types of reactions or interactions of one or more of the ingredients. Accordingly, the pharmaceutical compositions of the present invention encompass any composition made by admixing a compound of the present invention and a pharmaceutically acceptable carrier.

10

The terms "administration of" and "administering a" compound should be understood to mean providing a compound of the invention or a prodrug of a compound of the invention to the individual in need.

Another aspect of the present invention is concerned with a method of 15 inhibiting HCV NS5B polymerase, inhibiting HCV replication, or treating HCV infection with a compound of the present invention in combination with one or more agents useful for treating HCV infection. Such agents active against HCV include, but are not limited to, ribavirin, levovirin, viramidine, thymosin alpha-1, interferon- β , interferon- α , pegylated interferon- α (peginterferon- α), a combination of interferon- α 20 and ribavirin, a combination of peginterferon- α and ribavirin, a combination of

interferon- α and levovirin, and a combination of peginterferon- α and levovirin. Interferon- α includes, but is not limited to, recombinant interferon- α 2a (such as Roferon interferon available from Hoffmann-LaRoche, Nutley, NJ), pegylated interferon- α 2a (PegasysTM), interferon- α 2b (such as Intron-A interferon available from 5 Schering Corp., Kenilworth, NJ), pegylated interferon- α 2b (PegIntronTM), a recombinant consensus interferon (such as interferon alphacon-1), and a purified interferon- α product. Amgen's recombinant consensus interferon has the brand name Infergen®. Levovirin is the L-enantiomer of ribavirin which has shown 10 immunomodulatory activity similar to ribavirin. Viramidine represents an analog of ribavirin disclosed in WO 01/60379 (assigned to ICN Pharmaceuticals). In accordance with this method of the present invention, the individual components of the combination can be administered separately at different times during the course of therapy or concurrently in divided or single combination forms. The instant invention is therefore to be understood as embracing all such regimes of simultaneous or 15 alternating treatment, and the term "administering" is to be interpreted accordingly. It will be understood that the scope of combinations of the compounds of this invention with other agents useful for treating HCV infection includes in principle any combination with any pharmaceutical composition for treating HCV infection. When a compound of the present invention or a pharmaceutically acceptable salt thereof is 20 used in combination with a second therapeutic agent active against HCV, the dose of each compound may be either the same as or different from the dose when the compound is used alone.

For the treatment of HCV infection, the compounds of the present invention may also be administered in combination with an agent that is an inhibitor 25 of HCV NS3 serine protease. HCV NS3 serine protease is an essential viral enzyme and has been described to be an excellent target for inhibition of HCV replication. Both substrate and non-substrate based inhibitors of HCV NS3 protease inhibitors are disclosed in WO 98/22496, WO 98/46630, WO 99/07733, WO 99/07734, WO 99/38888, WO 99/50230, WO 99/64442, WO 00/09543, WO 00/59929, and GB-30 2337262. HCV NS3 protease as a target for the development of inhibitors of HCV replication and for the treatment of HCV infection is discussed in B.W. Dymock, "Emerging therapies for hepatitis C virus infection," Emerging Drugs, 6: 13-42 (2001).

Ribavirin, levovirin, and viramidine may exert their anti-HCV effects 35 by modulating intracellular pools of guanine nucleotides via inhibition of the

intracellular enzyme inosine monophosphate dehydrogenase (IMPDH). IMPDH is the rate-limiting enzyme on the biosynthetic route in *de novo* guanine nucleotide biosynthesis. Ribavirin is readily phosphorylated intracellularly and the monophosphate derivative is an inhibitor of IMPDH. Thus, inhibition of IMPDH

5 represents another useful target for the discovery of inhibitors of HCV replication. Therefore, the compounds of the present invention may also be administered in combination with an inhibitor of IMPDH, such as VX-497, which is disclosed in WO 97/41211 and WO 01/00622 (assigned to Vertex); another IMPDH inhibitor, such as that disclosed in WO 00/25780 (assigned to Bristol-Myers Squibb); or mycophenolate 10 mofetil [see A.C. Allison and E.M. Eugui, Agents Action, 44 (Suppl.): 165 (1993)].

For the treatment of HCV infection, the compounds of the present invention may also be administered in combination with the antiviral agent amantadine (1-aminoadamantane) [for a comprehensive description of this agent, see J. Kirschbaum, Anal. Profiles Drug Subs. 12: 1-36 (1983)].

15 The compounds of the present invention may also be combined for the treatment of HCV infection with antiviral 2'-C-branched ribonucleosides disclosed in R. E. Harry-O'kuru, et al., J. Org. Chem., 62: 1754-1759 (1997); M. S. Wolfe, et al., Tetrahedron Lett., 36: 7611-7614 (1995); U.S. Patent No. 3,480,613 (Nov. 25, 1969); International Publication Number WO 01/90121 (29 November 2001); International 20 Publication Number WO 01/92282 (6 December 2001); and International Publication Number WO 02/32920 (25 April 2002); the contents of each of which are incorporated by reference in their entirety. Such 2'-C-branched ribonucleosides include, but are not limited to, 2'-C-methyl-cytidine, 2'-C-methyl-uridine, 2'-C-methyl-adenosine, 2'-C-methyl-guanosine, and 9-(2-C-methyl- β -D-ribofuranosyl)-25 2,6-diaminopurine.

By "pharmaceutically acceptable" is meant that the carrier, diluent, or excipient must be compatible with the other ingredients of the formulation and not deleterious to the recipient thereof.

Also included within the present invention are pharmaceutical 30 compositions comprising the nucleoside compounds and derivatives thereof of the present invention in association with a pharmaceutically acceptable carrier. Another example of the invention is a pharmaceutical composition made by combining any of the compounds described above and a pharmaceutically acceptable carrier. Another illustration of the invention is a process for making a pharmaceutical composition

comprising combining any of the compounds described above and a pharmaceutically acceptable carrier.

Also included within the present invention are pharmaceutical compositions useful for inhibiting RNA-dependent RNA viral polymerase in particular HCV NS5B polymerase comprising an effective amount of a compound of the present invention and a pharmaceutically acceptable carrier. Pharmaceutical compositions useful for treating RNA-dependent RNA viral infection in particular HCV infection are also encompassed by the present invention as well as a method of inhibiting RNA-dependent RNA viral polymerase in particular HCV NS5B polymerase and a method of treating RNA-dependent viral replication and in particular HCV replication. Additionally, the present invention is directed to a pharmaceutical composition comprising a therapeutically effective amount of a compound of the present invention in combination with a therapeutically effective amount of another agent active against RNA-dependent RNA virus and in particular against HCV. Agents active against HCV include, but are not limited to, ribavirin, levovirin, viramidine, thymosin alpha-1, an inhibitor of HCV NS3 serine protease, interferon- α , pegylated interferon- α (peginterferon- α), a combination of interferon- α and ribavirin, a combination of peginterferon- α and ribavirin, a combination of interferon- α and levovirin, and a combination of peginterferon- α and levovirin.

Interferon- α includes, but is not limited to, recombinant interferon- α 2a (such as Roferon interferon available from Hoffmann-LaRoche, Nutley, NJ), interferon- α 2b (such as Intron-A interferon available from Schering Corp., Kenilworth, NJ), a consensus interferon, and a purified interferon- α product. For a discussion of ribavirin and its activity against HCV, see J.O. Saunders and S.A. Raybuck, "Inosine Monophosphate Dehydrogenase: Consideration of Structure, Kinetics, and Therapeutic Potential," Ann. Rep. Med. Chem., 35: 201-210 (2000).

Another aspect of the present invention provides for the use of the nucleoside compounds and derivatives thereof and their pharmaceutical compositions for the manufacture of a medicament for the inhibition of RNA-dependent RNA viral replication, in particular HCV replication, and/or the treatment of RNA-dependent RNA viral infection, in particular HCV infection. Yet a further aspect of the present invention provides for the nucleoside compounds and derivatives thereof and their pharmaceutical compositions for use as a medicament for the inhibition of RNA-dependent RNA viral replication, in particular HCV replication, and/or for the treatment of RNA-dependent RNA viral infection, in particular HCV infection.

The pharmaceutical compositions of the present invention comprise a compound of structural formula I as an active ingredient or a pharmaceutically acceptable salt thereof, and may also contain a pharmaceutically acceptable carrier and optionally other therapeutic ingredients.

5 The compositions include compositions suitable for oral, rectal, topical, parenteral (including subcutaneous, intramuscular, and intravenous), ocular (ophthalmic), pulmonary (nasal or buccal inhalation), or nasal administration, although the most suitable route in any given case will depend on the nature and severity of the conditions being treated and on the nature of the active ingredient.

10 They may be conveniently presented in unit dosage form and prepared by any of the methods well-known in the art of pharmacy.

In practical use, the compounds of structural formula I can be combined as the active ingredient in intimate admixture with a pharmaceutical carrier according to conventional pharmaceutical compounding techniques. The carrier may 15 take a wide variety of forms depending on the form of preparation desired for administration, e.g., oral or parenteral (including intravenous). In preparing the compositions for oral dosage form, any of the usual pharmaceutical media may be employed, such as, for example, water, glycols, oils, alcohols, flavoring agents, preservatives, coloring agents and the like in the case of oral liquid preparations, such 20 as, for example, suspensions, elixirs and solutions; or carriers such as starches, sugars, microcrystalline cellulose, diluents, granulating agents, lubricants, binders, disintegrating agents and the like in the case of oral solid preparations such as, for example, powders, hard and soft capsules and tablets, with the solid oral preparations being preferred over the liquid preparations.

25 Because of their ease of administration, tablets and capsules represent the most advantageous oral dosage unit form in which case solid pharmaceutical carriers are obviously employed. If desired, tablets may be coated by standard aqueous or nonaqueous techniques. Such compositions and preparations should contain at least 0.1 percent of active compound. The percentage of active compound 30 in these compositions may, of course, be varied and may conveniently be between about 2 percent to about 60 percent of the weight of the unit. The amount of active compound in such therapeutically useful compositions is such that an effective dosage will be obtained. The active compounds can also be administered intranasally as, for example, liquid drops or spray.

The tablets, pills, capsules, and the like may also contain a binder such as gum tragacanth, acacia, corn starch or gelatin; excipients such as dicalcium phosphate; a disintegrating agent such as corn starch, potato starch, alginic acid; a lubricant such as magnesium stearate; and a sweetening agent such as sucrose, lactose 5 or saccharin. When a dosage unit form is a capsule, it may contain, in addition to materials of the above type, a liquid carrier such as a fatty oil.

Various other materials may be present as coatings or to modify the physical form of the dosage unit. For instance, tablets may be coated with shellac, sugar or both. A syrup or elixir may contain, in addition to the active ingredient, 10 sucrose as a sweetening agent, methyl and propylparabens as preservatives, a dye and a flavoring such as cherry or orange flavor.

Compounds of structural formula I may also be administered parenterally. Solutions or suspensions of these active compounds can be prepared in water suitably mixed with a surfactant such as hydroxy-propylcellulose. Dispersions 15 can also be prepared in glycerol, liquid polyethylene glycols and mixtures thereof in oils. Under ordinary conditions of storage and use, these preparations contain a preservative to prevent the growth of microorganisms.

The pharmaceutical forms suitable for injectable use include sterile aqueous solutions or dispersions and sterile powders for the extemporaneous 20 preparation of sterile injectable solutions or dispersions. In all cases, the form must be sterile and must be fluid to the extent that easy syringability exists. It must be stable under the conditions of manufacture and storage and must be preserved against the contaminating action of microorganisms such as bacteria and fungi. The carrier can be a solvent or dispersion medium containing, for example, water, ethanol, polyol 25 (e.g. glycerol, propylene glycol and liquid polyethylene glycol), suitable mixtures thereof, and vegetable oils.

Any suitable route of administration may be employed for providing a mammal, especially a human with an effective dosage of a compound of the present 30 invention. For example, oral, rectal, topical, parenteral, ocular, pulmonary, nasal, and the like may be employed. Dosage forms include tablets, troches, dispersions, suspensions, solutions, capsules, creams, ointments, aerosols, and the like. Preferably compounds of structural formula I are administered orally.

For oral administration to humans, the dosage range is 0.01 to 1000 mg/kg body weight in divided doses. In one embodiment the dosage range is 0.1 to 35 100 mg/kg body weight in divided doses. In another embodiment the dosage range is

0.5 to 20 mg/kg body weight in divided doses. For oral administration, the compositions are preferably provided in the form of tablets or capsules containing 1.0 to 1000 milligrams of the active ingredient, particularly, 1, 5, 10, 15, 20, 25, 50, 75, 100, 150, 200, 250, 300, 400, 500, 600, 750, 800, 900, and 1000 milligrams of the

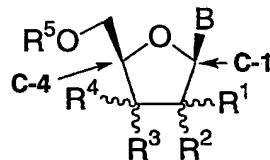
5 active ingredient for the symptomatic adjustment of the dosage to the patient to be treated.

The effective dosage of active ingredient employed may vary depending on the particular compound employed, the mode of administration, the condition being treated and the severity of the condition being treated. Such dosage

10 may be ascertained readily by a person skilled in the art. This dosage regimen may be adjusted to provide the optimal therapeutic response.

The compounds of the present invention contain one or more asymmetric centers and can thus occur as racemates and racemic mixtures, single enantiomers, diastereomeric mixtures and individual diastereomers. The present

15 invention is meant to comprehend nucleoside compounds having the β -D stereochemical configuration for the five-membered furanose ring as depicted in the structural formula below, that is, nucleoside compounds in which the substituents at C-1 and C-4 of the five-membered furanose ring have the β -stereochemical configuration ("up" orientation as denoted by a bold line).



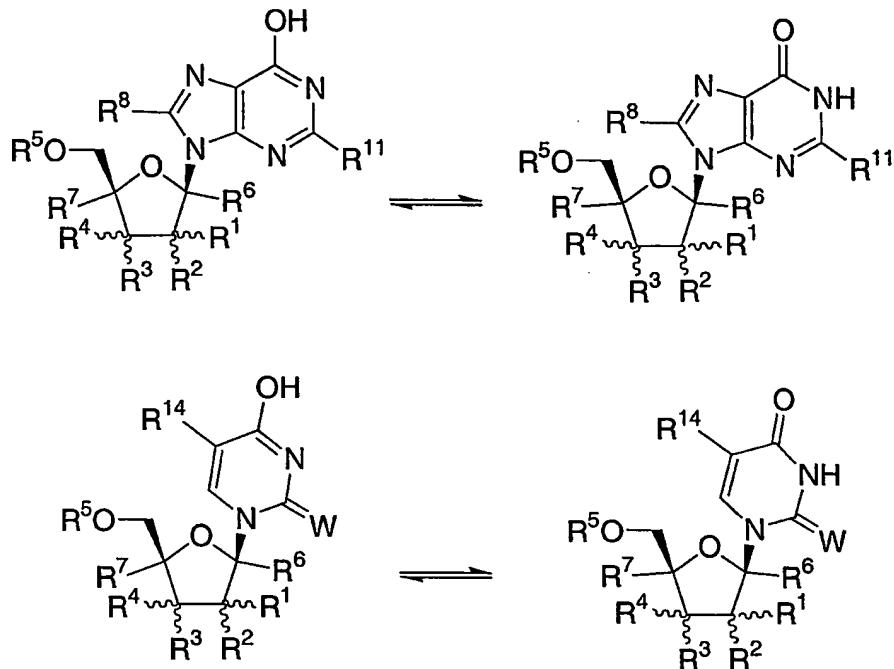
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 β -D-

Some of the compounds described herein contain olefinic double bonds, and unless specified otherwise, are meant to include both E and Z geometric isomers.

Some of the compounds described herein may exist as tautomers such

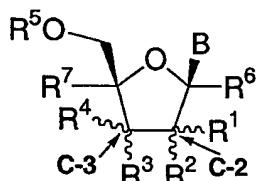
25 as keto-enol tautomers. The individual tautomers as well as mixtures thereof are encompassed with compounds of structural formula I. Examples of keto-enol tautomers which are intended to be encompassed within the compounds of the present invention are illustrated below:



Compounds of structural formula I may be separated into their individual diastereoisomers by, for example, fractional crystallization from a suitable solvent, for example methanol or ethyl acetate or a mixture thereof, or via chiral chromatography using an optically active stationary phase.

Alternatively, any stereoisomer of a compound of the structural formula I may be obtained by stereospecific synthesis using optically pure starting materials or reagents of known configuration.

The stereochemistry of the substituents at the C-2 and C-3 positions of the furanose ring of the compounds of the present invention of structural formula I is denoted by squiggly lines which signifies that substituents R1, R2, R3 and R4 can have either the α (substituent "down") or β (substituent "up") configuration independently of one another. Notation of stereochemistry by a bold line as at C-1 and C-4 of the furanose ring signifies that the substituent has the β -configuration (substituent "up").



(I)

The compounds of the present invention may be administered in the form of a pharmaceutically acceptable salt. The term "pharmaceutically acceptable salt" refers to salts prepared from pharmaceutically acceptable non-toxic bases or acids including inorganic or organic bases and inorganic or organic acids. Salts of basic compounds encompassed within the term "pharmaceutically acceptable salt" refer to non-toxic salts of the compounds of this invention which are generally prepared by reacting the free base with a suitable organic or inorganic acid.

Representative salts of basic compounds of the present invention include, but are not limited to, the following: acetate, benzenesulfonate, benzoate, bicarbonate, bisulfate, bitartrate, borate, bromide, camsylate, carbonate, chloride, clavulanate, citrate, dihydrochloride, edetate, edisylate, estolate, esylate, fumarate, gluceptate, gluconate, glutamate, glycolylarsanilate, hexylresorcinate, hydrabamine, hydrobromide, hydrochloride, hydroxynaphthoate, iodide, isothionate, lactate, lactobionate, laurate, malate, maleate, mandelate, mesylate, methylbromide, methylnitrate, methylsulfate, mucate, napsylate, nitrate, N-methylglucamine ammonium salt, oleate, oxalate, pamoate (embonate), palmitate, pantothenate, phosphate/diphosphate, polygalacturonate, salicylate, stearate, sulfate, subacetate, succinate, tannate, tartrate, teoclolate, tosylate, triethiodide and valerate. Furthermore, where the compounds of the invention carry an acidic moiety, suitable pharmaceutically acceptable salts thereof include, but are not limited to, salts derived from inorganic bases including aluminum, ammonium, calcium, copper, ferric, ferrous, lithium, magnesium, manganese, mangamous, potassium, sodium, zinc, and the like. Particularly preferred are the ammonium, calcium, magnesium, potassium, and sodium salts. Salts derived from pharmaceutically acceptable organic non-toxic bases include salts of primary, secondary, and tertiary amines, cyclic amines, and basic ion-exchange resins, such as arginine, betaine, caffeine, choline, N,N-dibenzylethylenediamine, diethylamine, 2-diethylaminoethanol, 2-dimethylaminoethanol, ethanolamine, ethylenediamine, N-ethylmorpholine, N-ethylpiperidine, glucamine, glucosamine, histidine, hydrabamine,

isopropylamine, lysine, methylglucamine, morpholine, piperazine, piperidine, polyamine resins, procaine, purines, theobromine, triethylamine, trimethylamine, tripropylamine, tromethamine, and the like.

Also, in the case of a carboxylic acid (-COOH) or alcohol group being 5 present in the compounds of the present invention, pharmaceutically acceptable esters of carboxylic acid derivatives, such as methyl, ethyl, or pivaloyloxymethyl, or acyl derivatives of alcohols, such as acetate, octanoate, or maleate, can be employed. Included are those esters and acyl groups known in the art for modifying the solubility or hydrolysis characteristics for use as sustained-release or prodrug formulations.

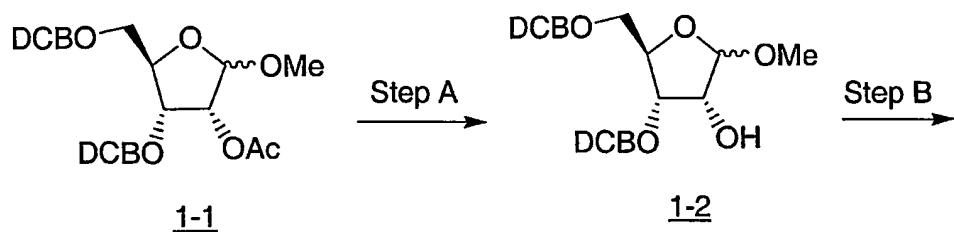
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Preparation of the Nucleoside Compounds and Derivatives of the Invention

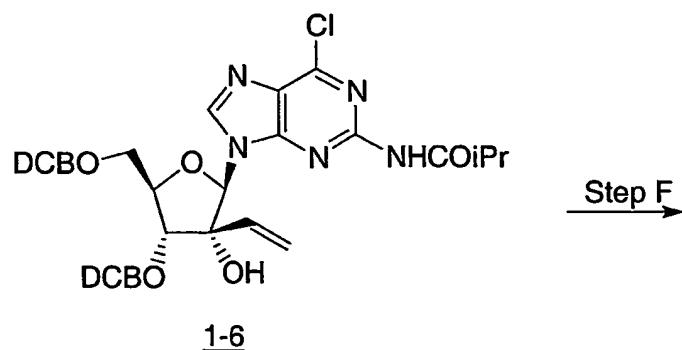
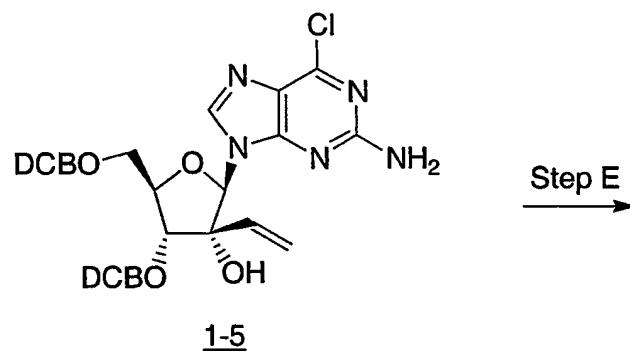
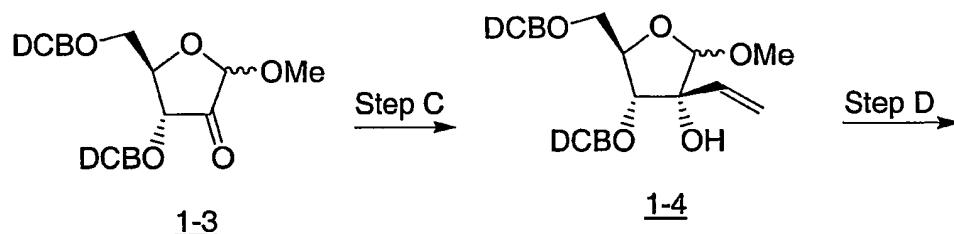
The nucleoside compounds and derivatives thereof of the present invention can be prepared following synthetic methodologies well-established in the practice of nucleoside and nucleotide chemistry. Reference is made to the following 15 text for a description of synthetic methods used in the preparation of the compounds of the present invention: "Chemistry of Nucleosides and Nucleotides," L.B. Townsend, ed., Vols. 1-3, Plenum Press, 1988, which is incorporated by reference herein in its entirety.

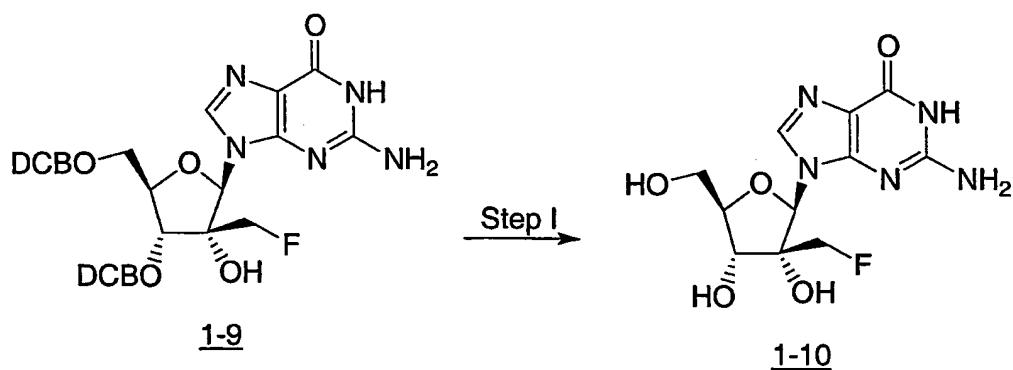
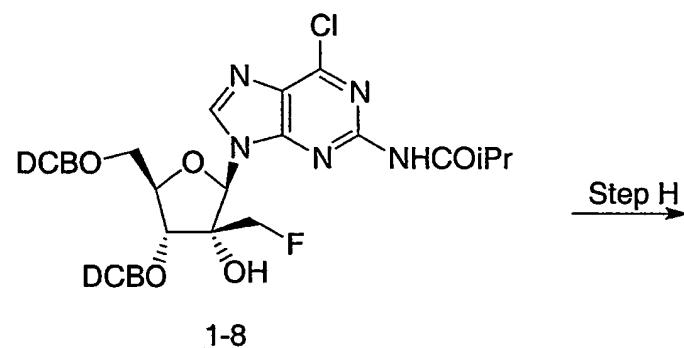
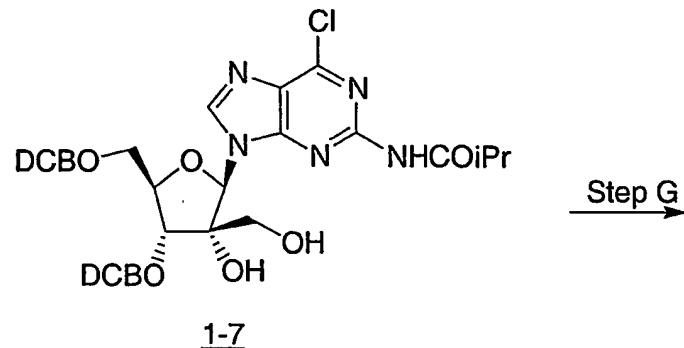
The examples below provide citations to literature publications, which 20 contain details for the preparation of final compounds or intermediates employed in the preparation of final compounds of the present invention. The nucleoside compounds of the present invention were prepared according to procedures detailed in the following examples. The examples are not intended to be limitations on the scope of the instant invention in any way, and they should not be so construed. Those 25 skilled in the art of nucleoside and nucleotide synthesis will readily appreciate that known variations of the conditions and processes of the following preparative procedures can be used to prepare these and other compounds of the present invention. All temperatures are degrees Celsius unless otherwise noted.

Scheme 1



(DCB = 2,4-dichlorobenzyl)



2-Amino-9-(2-C-fluoromethyl-β-D-ribofuranosyl)-3,9-dihydropurin-6-one (1-10)

Step A: 3,5-Bis-O-(2,4-dichlorobenzyl)-1-O-methyl-α-D-ribofuranose (1-2)

A mixture of 2-*O*-acetyl-3,5-bis-*O*-(2,4-dichlorobenzyl)-1-*O*-methyl- α -D-ribofuranose (1-1) [for preparation, see: *Helv. Chim. Acta* 78: 486 (1995)] (52.4 g, 0.10 mol) in methanolic K₂CO₃ (500 mL, saturated at room temperature) was stirred at room temperature for 45 min. and then concentrated under reduced pressure. The 5 oily residue was suspended in CH₂Cl₂ (500 mL), washed with water (300 mL + 5 \times 200 mL) and brine (200 mL), dried (Na₂SO₄), filtered, and concentrated to give the title compound (49.0 g) as colorless oil, which was used without further purification in Step B below.

10 ¹H NMR (DMSO-*d*₆): δ 3.28 (s, 3H, OCH₃), 3.53 (d, 2H, *J*_{5,4} = 4.5 Hz, H-5a, H-5b), 3.72 (dd, 1H, *J*_{3,4} = 3.6 Hz, *J*_{3,2} = 6.6 Hz, H-3), 3.99 (ddd, 1H, *J*_{2,1} = 4.5 Hz, *J*_{2,OH-2} = 9.6 Hz, H-2), 4.07 (m, 1H, H-4), 4.50 (s, 2H, CH₂Ph), 4.52, 4.60 (2d, 2H, *J*_{gem} = 13.6 Hz, CH₂Ph), 4.54 (d, 1H, OH-2), 4.75 (d, 1H, H-1), 7.32-7.45, 7.52-7.57 (2m, 10H, 2Ph).

15 ¹³C NMR (DMSO-*d*₆): δ 55.40, 69.05, 69.74, 71.29, 72.02, 78.41, 81.45, 103.44, 127.83, 127.95, 129.05, 129.28, 131.27, 131.30, 133.22, 133.26, 133.55, 133.67, 135.45, 135.92.

Step B: 3,5-Bis-*O*-(2,4-dichlorobenzyl)-1-*O*-methyl- α -D-*erythro*-pentofuranos-2-ulose (1-3)

20 To an ice-cold suspension of Dess-Martin periodinane (50.0 g, 118 mmol) in anhydrous CH₂Cl₂ (350 mL) under argon (Ar) was added a solution of the compound from Step A (36.2 g, 75 mmol) in anhydrous CH₂Cl₂ (200 mL) dropwise over 0.5 h. The reaction mixture was stirred at 0°C for 0.5 h and then at room temperature for 3 days. The mixture was diluted with anhydrous Et₂O (600 mL) and 25 poured into an ice-cold mixture of Na₂S₂O₃.5H₂O (180 g) in saturated aqueous NaHCO₃ (1400 mL). The layers were separated, and the organic layer was washed with saturated aqueous NaHCO₃ (600 mL), water (800 mL) and brine (600 mL), dried (MgSO₄), filtered and evaporated to give the title compound (34.2 g) as a colorless oil, which was used without further purification in Step C below.

30 ¹H NMR (CDCl₃): δ 3.50 (s, 3H, OCH₃), 3.79 (dd, 1H, *J*_{5a,5b} = 11.3 Hz, *J*_{5a,4} = 3.5 Hz, H-5a), 3.94 (dd, 1H, *J*_{5b,4} = 2.3 Hz, H-5b), 4.20 (dd, 1H, *J*_{3,1} = 1.3 Hz, *J*_{3,4} = 8.4 Hz, H-3), 4.37 (ddd, 1H, H-4), 4.58, 4.69 (2d, 2H, *J*_{gem} = 13.0 Hz, CH₂Ph), 4.87 (d, 1H, H-1), 4.78, 5.03 (2d, 2H, *J*_{gem} = 12.5 Hz, CH₂Ph), 7.19-7.26, 7.31-7.42 (2m, 10H, 2Ph).

¹³C NMR (DMSO-*d*₆): δ 55.72, 69.41, 69.81, 69.98, 77.49, 78.00, 98.54, 127.99, 128.06, 129.33, 129.38, 131.36, 131.72, 133.61, 133.63, 133.85, 133.97, 134.72, 135.32, 208.21.

5 Step C: 3,5-Bis-*O*-(2,4-dichlorobenzyl)-2-C-vinyl-1-*O*-methyl- α -D-ribofuranose (1-4)

Cerium chloride heptahydrate (50 g, 134.2 mmol) was finely crushed in a pre-heated mortar and transferred to a round-bottom flask equipped with a mechanical stirrer. The flask was heated under high vacuum overnight at 160°C. The 10 vacuum was released under argon and the flask was cooled to room temperature. Anhydrous THF (300 mL) was cannulated into the flask. The resulting suspension was stirred at room temperature for 4 h and then cooled to -78 °C. Vinylmagnesium bromide (1M in THF, 120 mL, 120 mmol) was added and stirring continued at -78 °C for 2 h. To this suspension was added a solution of 3,5-bis-*O*-(2,4-15 dichlorophenylmethyl)-1-*O*-methyl- α -D-erythro-pentofuranose-2-ulose (14 g, 30 mmol) [from Example 2, Step B] in anhydrous THF (100 mL), dropwise with constant stirring. The reaction was stirred at -78 °C for 4 h. The reaction was quenched with saturated ammonium chloride solution and allowed to come to room temperature. The mixture was filtered through a celite pad and the residue washed 20 with Et₂O (2 × 500 mL). The organic layer was separated and the aqueous layer extracted with Et₂O (2 × 200 mL). The combined organic layers were dried over anhydrous Na₂SO₄ and concentrated to a viscous yellow oil. The oil was purified by flash chromatography (SiO₂, 10% EtOAc in hexanes). The title compound (6.7 g, 13.2 mmol) was obtained as a pale yellow oil.

25

Step D: 2-Amino-6-chloro-9-[3,5-bis-*O*-(2,4-dichlorobenzyl)-2-C-vinyl- β -D-ribofuranosyl]-9H-purine (1-5)

To a solution of compound from Step A (1-4) (6.4 g, 12.6 mmol) in anhydrous dichloromethane (150 mL) at -20 °C is added dropwise 30%HBr in AcOH 30 (20 mL, 75.6 mmol). The resulting solution is stirred between -10 °C and 0 °C for 4 h, evaporated in vacuo and coevaporated with anhydrous toluene (3 × 40 mL). The oily residue is dissolved in anhydrous acetonitrile (100 mL) and added to a solution of sodium salt of 2-amino-6-chloro-9H-purine (3 eq) in acetonitrile (generated in situ from 2-amino-6-chloro-9H-purine (3 eq) and sodium hydride (3 eq) at -20 °C. The

resulting mixture is allowed to come to room temperature and stirred at room temperature for 24 h. The mixture is evaporated to dryness, taken up in water and extracted with EtOAc (2×300 mL). The combined organic extracts are dried over Na_2SO_4 , filtered and evaporated. The crude product is purified by flash

5 chromatography on silica gel eluting with 10% EtOAc in hexanes to afford the title compound 1-5.

Step E: 2-(Isobutyrylamino)-6-chloro-9-[3,5-bis-O-(2,4-dichlorobenzyl)-2-C-vinyl-β-D-ribofuranosyl]-9H-purine (1-6)

10 The compound from Step D (1-5) is dissolved in anhydrous pyridine and cooled to 0 °C. A solution of isobutyryl chloride (1.5 eq) in anhydrous dichloromethane is added and the reaction stirred at room temperature for 24 h under argon. The reaction mixture is concentrated and the residue subjected to silica gel chromatography to give the title compound 1-6.

15

Step F: 2-(Isobutyrylamino)-6-chloro-9-[3,5-bis-O-(2,4-dichlorobenzyl)-2-C-hydroxymethyl-β-D-ribofuranosyl]-9H-purine (1-7)

To a solution of the compound from Step E in 1,4-dioxane is added N-methylmorpholine-N-oxide (3 eq) and osmium tetroxide (4% solution in water, 0.1

20 eq). The mixture is stirred in the dark for 14 h. The precipitate is removed by filtration through a filter aid, diluted with water and extracted with EtOAc. The EtOAc layer is dried over Na_2SO_4 and concentrated in vacuo. The oily residue is taken up in dichloromethane and stirred over NaIO_4 on silica gel (1 gm/ 100 mg of starting material) for 12 h. The silica gel is removed by filtration and the residue is
25 evaporated and taken up in absolute ethanol. The solution is cooled in an ice bath and sodium borohydride (5 eq) added in small portions. The resulting mixture is stirred at room temperature for 4 h and then diluted with EtOAc. The organic layer is washed with water, brine and dried over Na_2SO_4 . The solvent is removed by evaporation and the residue purified by flash chromatography on silica gel to give the title compound
30 1-7.

Step G: 2-(Isobutyrylamino)-6-chloro-9-[3,5-bis-O-(2,4-dichlorobenzyl)-2-C-fluoromethyl-β-D-ribofuranosyl]-9H-purine (1-8)

35 To a solution of the compound from Step F in anhydrous dichloromethane under argon is added 4-dimethylaminopyridine (0.1 eq) and

triethylamine (3 eq). The solution is cooled to -10 °C and trifluoromethanesulfonic anhydride (1.2 eq) is added. The reaction is stirred in the cold for 1 h, washed with ice-cold saturated NaHCO₃ solution, ice water, ice-cold brine, dried over Na₂SO₄ and concentrated in vacuo. The residue is dissolved in anhydrous THF (5 mL) and cooled 5 in an icebath. Tetrabutylammonium fluoride (1M solution in THF, 10 eq) is added and the mixture stirred at room temperature for 4 h. The solvent is removed by evaporation under diminished pressure, the residue taken up in dichloromethane, and washed with saturated NaHCO₃ solution, water and brine. The dichloromethane layer is dried over anhydrous Na₂SO₄, concentrated in vacuo, and the residue subjected to 10 chromatography on silica gel to afford the title compound 1-8.

Step H: 2-Amino-9-[3,5-bis-O-(2,4-dichlorobenzyl)-2-C-fluoromethyl-β-D-ribofuranosyl]-3,9-dihydropurin-6-one (1-9)

The compound from Step G is dissolved in the minimum amount of 15 1,4-dioxane. 4N NaOH (equivalent volume to that of 1,4-dioxane) is added and the reaction refluxed for 4 h. The reaction is cooled to room temperature and neutralized with 4N HCl. The solvent is removed by evaporation under diminished pressure and the residue applied to a silica gel column. The title compound is eluted with a combination of methanol and dichloromethane. The appropriate fractions are 20 combined and evaporated under diminished pressure to afford the title compound 1-9.

Step I: 2-Amino-9-(2-C-fluoromethyl-β-D-ribofuranosyl)-3,9-dihydropurin-6-one (1-10)

The compound from Step H is taken up in 1:1 methanol 25 /dichloromethane and the mixture is hydrogenated in the presence of 10% Pd/C (1 eq by weight) for 12 h. The catalyst is removed by filtration through a filter aid and the filter washed with copious amounts of methanol. The combined filtrate and washings are evaporated in vacuo and the residue purified by silica gel chromatography to give the title compound 1-10.

30

BIOLOGICAL ASSAYS

The assays employed to measure the inhibition of HCV NS5B polymerase and HCV replication are described below.

The effectiveness of the compounds of the present invention as inhibitors of HCV NS5B RNA-dependent RNA polymerase (RdRp) was measured in the following assay.

A. Assay for Inhibition of HCV NS5B Polymerase:

5 This assay was used to measure the ability of the nucleoside derivatives of the present invention to inhibit the enzymatic activity of the RNA-dependent RNA polymerase (NS5B) of the hepatitis C virus (HCV) on a heteromeric RNA template.

10 Procedure:

Assay Buffer Conditions: (50 μ L -total/reaction)

20 mM Tris, pH 7.5

50 μ M EDTA

5 mM DTT

15 2 mM MgCl₂

80 mM KCl

0.4 U/ μ L RNasin (Promega, stock is 40 units/ μ L)

0.75 μ g t500 (a 500-nt RNA made using T7 runoff transcription with a sequence from the NS2/3 region of the hepatitis C genome)

20 1.6 μ g purified hepatitis C NS5B (form with 21 amino acids C-terminally truncated)

1 μ M A,C,U,GTP (Nucleoside triphosphate mix)

[alpha-³²P]-GTP or [alpha-³³P]-GTP

25 The compounds were tested at various concentrations up to 100 μ M final concentration.

An appropriate volume of reaction buffer was made including enzyme and template t500. Nucleoside derivatives of the present invention were pipetted into the wells of a 96-well plate. A mixture of nucleoside triphosphates (NTP's), including the radiolabeled GTP, was made and pipetted into the wells of a 96-well plate. The reaction was initiated by addition of the enzyme-template reaction solution and allowed to proceed at room temperature for 1-2 h.

30 The reaction was quenched by addition of 20 μ L 0.5M EDTA, pH 8.0. Blank reactions in which the quench solution was added to the NTPs prior to the addition of the reaction buffer were included.

50 μ L of the quenched reaction were spotted onto DE81 filter disks (Whatman) and allowed to dry for 30 min. The filters were washed with 0.3 M ammonium formate, pH 8 (150 mL/wash until the cpm in 1 mL wash is less than 100, usually 6 washes). The filters were counted in 5-mL scintillation fluid in a scintillation counter.

5 The percentage of inhibition was calculated according to the following equation: %Inhibition = [1-(cpm in test reaction - cpm in blank) / (cpm in control reaction - cpm in blank)] x 100.

10 Representative compounds tested in the HCV NS5B polymerase assay exhibited IC₅₀'s less than 100 micromolar.

B. Assay for Inhibition of HCV RNA Replication:

15 The compounds of the present invention were also evaluated for their ability to affect the replication of Hepatitis C Virus RNA in cultured hepatoma (HuH-7) cells containing a subgenomic HCV Replicon. The details of the assay are described below. This Replicon assay is a modification of that described in V. Lohmann, F. Korner, J-O. Koch, U. Herian, L. Theilmann, and R. Bartenschlager, "Replication of a Sub-genomic Hepatitis C Virus RNAs in a Hepatoma Cell Line," Science 285:110 (1999).

20

Protocol:

25 The assay was an *in situ* Ribonuclease protection, Scintillation Proximity based-plate assay (SPA). 10,000 - 40,000 cells were plated in 100-200 μ L of media containing 0.8mg/mL G418 in 96-well cytosar plates (Amersham). Compounds were added to cells at various concentrations up to 100 μ M in 1% DMSO at time 0 to 18 h and then cultured for 24-96 h. Cells were fixed (20 min, 10% formalin), permeabilized (20 min, 0.25% Triton X-100/PBS) and hybridized (overnight, 50°C) with a single-stranded ³³P RNA probe complementary to the (+) strand NS5B (or other genes) contained in the RNA viral genome. Cells were washed, treated with RNase, washed, heated to 65°C and counted in a Top-Count. Inhibition of replication was read as a decrease in counts per minute (cpm).

30 Human HuH-7 hepatoma cells, which were selected to contain a subgenomic replicon, carry a cytoplasmic RNA consisting of an HCV 5' non-translated region (NTR), a neomycin selectable marker, an EMCV IRES (internal

ribosome entry site), and HCV non-structural proteins NS3 through NS5B, followed by the 3' NTR.

Representative compounds tested in the replication assay exhibited EC₅₀'s less than 100 micromolar.

5

The nucleoside derivatives of the present invention were also evaluated for cellular toxicity and anti-viral specificity in the counterscreens described below.

C. COUNTERSCREENS:

10 The ability of the nucleoside derivatives of the present invention to inhibit human DNA polymerases was measured in the following assays.

a. Inhibition of Human DNA Polymerases alpha and beta:

15 Reaction Conditions:

50 μ L reaction volume

Reaction buffer components:

20 mM Tris-HCl, pH 7.5

200 μ g/mL bovine serum albumin

100 mM KCl

2 mM β -mercaptoethanol

10 mM MgCl₂

1.6 μ M dA, dG, dC, dTTP

25 α -³²P-dATP

Enzyme and template:

0.05 mg/mL gapped fish sperm DNA template

0.01 U/ μ L DNA polymerase α or β

30

Preparation of gapped fish sperm DNA template:

Add 5 μ L 1M MgCl₂ to 500 μ L activated fish sperm DNA (USB 70076);

Warm to 37°C and add 30 μ L of 65 U/ μ L of exonuclease III (GibcoBRL 18013-011);

Incubate 5 min at 37°C;

35 Terminate reaction by heating to 65 °C for 10 min;

Load 50-100 μ L aliquots onto Bio-spin 6 chromatography columns (Bio-Rad 732-6002) equilibrated with 20 mM Tris-HCl, pH 7.5;
Elute by centrifugation at 1,000Xg for 4 min;
Pool eluate and measure absorbance at 260 nm to determine concentration.

5

The DNA template was diluted into an appropriate volume of 20 mM Tris-HCl, pH 7.5 and the enzyme was diluted into an appropriate volume of 20 mM Tris-HCl, containing 2 mM β -mercaptoethanol, and 100 mM KCl. Template and enzyme were pipetted into microcentrifuge tubes or a 96 well plate. Blank reactions 10 excluding enzyme and control reactions excluding test compound were also prepared using enzyme dilution buffer and test compound solvent, respectively. The reaction was initiated with reaction buffer with components as listed above. The reaction was incubated for 1 hour at 37°C. The reaction was quenched by the addition of 20 μ L 0.5M EDTA. 50 μ L of the quenched reaction was spotted onto Whatman DE81 filter 15 disks and air dried. The filter disks were repeatedly washed with 150 mL 0.3M ammonium formate, pH 8 until 1 mL of wash is < 100 cpm. The disks were washed twice with 150 mL absolute ethanol and once with 150 mL anhydrous ether, dried and counted in 5 mL scintillation fluid.

The percentage of inhibition was calculated according to the following 20 equation: % inhibition = [1-(cpm in test reaction - cpm in blank)/(cpm in control reaction - cpm in blank)] x 100.

b. Inhibition of Human DNA Polymerase gamma :

The potential for inhibition of human DNA polymerase gamma was 25 measured in reactions that included 0.5 ng/ μ L enzyme; 10 μ M dATP, dGTP, dCTP, and TTP; 2 μ Ci/reaction [α -³³P]-dATP, and 0.4 μ g/ μ L activated fish sperm DNA (purchased from US Biochemical) in a buffer containing 20 mM Tris pH8, 2 mM β -mercaptoethanol, 50 mM KCl, 10 mM MgCl₂, and 0.1 μ g/ μ L BSA. Reactions were allowed to proceed for 1 h at 37°C and were quenched by addition of 0.5 M EDTA to 30 a final concentration of 142 mM. Product formation was quantified by anion exchange filter binding and scintillation counting. Compounds were tested at up to 50 μ M.

The percentage of inhibition was calculated according to the following 35 equation: % inhibition = [1-(cpm in test reaction - cpm in blank)/(cpm in control reaction - cpm in blank)] x 100.

The ability of the nucleoside derivatives of the present invention to inhibit HIV infectivity and HIV spread was measured in the following assays.

5 c. HIV Infectivity Assay

Assays were performed with a variant of HeLa Magi cells expressing both CXCR4 and CCR5 selected for low background β -galactosidase (β -gal) expression. Cells were infected for 48 h, and β -gal production from the integrated HIV-1 LTR promoter was quantified with a chemiluminescent substrate (Galactolight 10 Plus, Tropix, Bedford, MA). Inhibitors were titrated (in duplicate) in twofold serial dilutions starting at 100 μ M; percent inhibition at each concentration was calculated in relation to the control infection.

15 d. Inhibition of HIV Spread

The ability of the compounds of the present invention to inhibit the spread of the human immunodeficiency virus (HIV) was measured by the method described in U.S. Patent No. 5,413,999 (May 9, 1995), and J.P. Vacca, et al., Proc. Natl. Acad. Sci., 91: 4096-4100 (1994), which are incorporated by reference herein in their entirety.

20

The nucleoside derivatives of the present invention were also screened for cytotoxicity against cultured hepatoma (HuH-7) cells containing a subgenomic HCV Replicon in an MTS cell-based assay as described in the assay below. The HuH-7 cell line is described in H. Nakabayashi, et al., Cancer Res., 42: 3858 (1982).

25

e. Cytotoxicity assay:

Cell cultures were prepared in appropriate media at concentrations of approximately 1.5×10^5 cells/mL for suspension cultures in 3 day incubations and 5.0×10^4 cells/mL for adherent cultures in 3 day incubations. 99 μ L of cell culture was transferred to wells of a 96-well tissue culture treated plate, and 1 μ L of 100-times final concentration of the test compound in DMSO was added. The plates were incubated at 37°C and 5% CO₂ for a specified period of time. After the incubation period, 20 μ L of CellTiter 96 Aqueous One Solution Cell Proliferation Assay reagent (MTS) (Promega) was added to each well and the plates were incubated at 37°C and 35 5% CO₂ for an additional period of time up to 3 h. The plates were agitated to mix

well and absorbance at 490 nm was read using a plate reader. A standard curve of suspension culture cells was prepared with known cell numbers just prior to the addition of MTS reagent. Metabolically active cells reduce MTS to formazan. Formazan absorbs at 490 nm. The absorbance at 490 nm in the presence of 5 compound was compared to absorbance in cells without any compound added. Reference: Cory, A. H. *et al.*, "Use of an aqueous soluble tetrazolium/formazan assay for cell growth assays in culture," Cancer Commun. 3: 207 (1991).

10 The following assays were employed to measure the activity of the compounds of the present invention against other RNA-dependent RNA viruses:

a. Determination of In Vitro Antiviral Activity of Compounds Against Rhinovirus (Cytopathic Effect Inhibition Assay):

15 Assay conditions are described in the article by Sidwell and Huffman, "Use of disposable microtissue culture plates for antiviral and interferon induction studies," Appl. Microbiol. 22: 797-801 (1971).

Viruses:

20 Rhinovirus type 2 (RV-2), strain HGP, was used with KB cells and media (0.1% NaHCO₃, no antibiotics) as stated in the Sidwell and Huffman reference. The virus, obtained from the ATCC, was from a throat swab of an adult male with a mild acute febrile upper respiratory illness.

25 Rhinovirus type 9 (RV-9), strain 211, and rhinovirus type 14 (RV-14), strain Tow, were also obtained from the American Type Culture Collection (ATCC) in Rockville, MD. RV-9 was from human throat washings and RV-14 was from a throat swab of a young adult with upper respiratory illness. Both of these viruses were used with HeLa Ohio-1 cells (Dr. Fred Hayden, Univ. of VA) which were human cervical epitheloid carcinoma cells. MEM (Eagle's minimum essential medium) with 5% Fetal Bovine serum (FBS) and 0.1% NaHCO₃ was used as the growth medium.

30 Antiviral test medium for all three virus types was MEM with 5% FBS, 0.1% NaHCO₃, 50 µg gentamicin/mL, and 10 mM MgCl₂. 2000 µg/mL was the highest concentration used to assay the compounds of the present invention. Virus was added to the assay plate approximately 5 min after the test compound. Proper controls were also run. Assay plates were incubated with 35 humidified air and 5% CO₂ at 37°C. Cytotoxicity was monitored in the control cells

microscopically for morphologic changes. Regression analysis of the virus CPE data and the toxicity control data gave the ED50 (50% effective dose) and CC50 (50% cytotoxic concentration). The selectivity index (SI) was calculated by the formula: SI = CC50 ÷ ED50.

5

b. Determination of In Vitro Antiviral Activity of Compounds Against Dengue, Banzi, and Yellow Fever (CPE Inhibition Assay)

Assay details are provided in the Sidwell and Huffman reference above.

Viruses:

- 10 Dengue virus type 2, New Guinea strain, was obtained from the Center for Disease Control. Two lines of African green monkey kidney cells were used to culture the virus (Vero) and to perform antiviral testing (MA-104). Both Yellow fever virus, 17D strain, prepared from infected mouse brain, and Banzi virus, H 336 strain, isolated from the serum of a febrile boy in South Africa, were obtained from ATCC. Vero cells were used with both of these viruses and for assay.
- 15

Cells and Media:

MA-104 cells (BioWhittaker, Inc., Walkersville, MD) and Vero cells (ATCC) were used in Medium 199 with 5% FBS and 0.1% NaHCO₃ and without antibiotics.

- 20 Assay medium for dengue, yellow fever, and Banzi viruses was MEM, 2% FBS, 0.18% NaHCO₃ and 50 µg gentamicin/mL.

- 25 Antiviral testing of the compounds of the present invention was performed according to the Sidwell and Huffman reference and similar to the above rhinovirus antiviral testing. Adequate cytopathic effect (CPE) readings were achieved after 5-6 days for each of these viruses.

c. Determination of In Vitro Antiviral Activity of Compounds Against West Nile Virus (CPE Inhibition Assay)

- 30 Assay details are provided in the Sidwell and Huffman reference cited above. West Nile virus, New York isolate derived from crow brain, was obtained from the Center for Disease Control. Vero cells were grown and used as described above. Test medium was MEM, 1% FBS, 0.1% NaHCO₃ and 50 µg gentamicin/mL.

Antiviral testing of the compounds of the present invention was performed following the methods of Sidwell and Huffman which are similar to those used to assay for rhinovirus activity. Adequate cytopathic effect (CPE) readings were achieved after 5-6 days.

5

d. Determination of In Vitro Antiviral Activity of Compounds Against rhino, yellow fever, dengue, Banzi, and West Nile Viruses (Neutral Red Uptake Assay)

After performing the CPE inhibition assays above, an additional cytopathic detection method was used which is described in "Microtiter Assay for 10 Interferon: Microspectrophotometric Quantitation of Cytopathic Effect," Appl. Environ. Microbiol. 31: 35-38 (1976). A Model EL309 microplate reader (Bio-Tek Instruments Inc.) was used to read the assay plate. ED50's and CD50's were calculated as above.

15

EXAMPLE OF A PHARMACEUTICAL FORMULATION

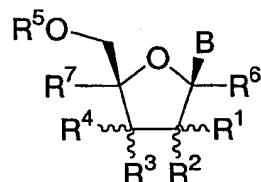
As a specific embodiment of an oral composition of a compound of the present invention, 50 mg of the compound of Example 1 is formulated with sufficient finely divided lactose to provide a total amount of 580 to 590 mg to fill a size O hard gelatin capsule.

20

While the invention has been described and illustrated in reference to specific embodiments thereof, those skilled in the art will appreciate that various changes, modifications, and substitutions can be made therein without departing from the spirit and scope of the invention. For example, effective dosages other than the 25 preferred doses as set forth hereinabove may be applicable as a consequence of variations in the responsiveness of the human being treated for severity of the HCV infection. Likewise, the pharmacologic response observed may vary according to and depending upon the particular active compound selected or whether there are present pharmaceutical carriers, as well as the type of formulation and mode of administration 30 employed, and such expected variations or differences in the results are contemplated in accordance with the objects and practices of the present invention. It is intended therefore that the invention be limited only by the scope of the claims which follow and that such claims be interpreted as broadly as is reasonable.

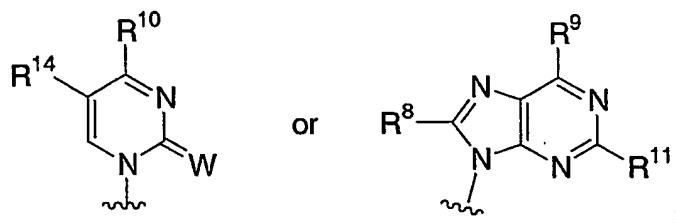
WHAT IS CLAIMED IS:

1. A compound of structural formula I:



(I)

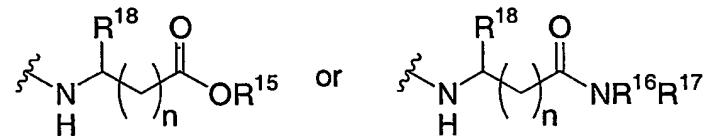
5 or a pharmaceutically acceptable salt thereof;
wherein B is



W is O or S;

10 R1 is fluoromethyl, difluoromethyl, or trifluoromethyl;
R2 is hydrogen, fluorine, amino, hydroxy, mercapto, C1-4 alkoxy, C1-8 alkylcarbonyloxy, or C1-4 alkyl;
R3 and R4 are each independently selected from the group consisting of hydrogen, cyano, azido, halogen, hydroxy, mercapto, amino, C1-4 alkoxy, C1-8 alkylcarbonyloxy, C2-4 alkenyl, C2-4 alkynyl, and C1-4 alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C1-4 alkoxy, C1-4 alkylthio, or one to three fluorine atoms;
R5 is hydrogen, C1-10 alkylcarbonyl, P3O9H4, P2O6H3, or P(O)R12R13;
R6 and R7 are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl;
20 R8 is hydrogen, C1-4 alkyl, C2-4 alkynyl, halogen, cyano, carboxy, C1-4 alkyloxycarbonyl, azido, amino, C1-4 alkylamino, di(C1-4 alkyl)amino, hydroxy, C1-6 alkoxy, C1-6 alkylthio, C1-6 alkylsulfonyl, or (C1-4 alkyl)0-2 aminomethyl;

R9 and R10 are each independently hydrogen, hydroxy, mercapto, halogen, C1-4 alkoxy, C1-4 alkylthio, C1-8 alkylcarbonyloxy, C3-6 cycloalkylcarbonyloxy, C1-8 alkyloxycarbonyloxy, C3-6 cycloalkyloxycarbonyloxy, -OCH₂CH₂SC(=O)C1-4 alkyl, -OCH₂O(C=O)C1-4 alkyl, -OCH(C1-4 alkyl)O(C=O)C1-4 alkyl, amino, C1-4 alkylamino, di(C1-4 alkyl)amino, C3-6 cycloalkylamino, di(C3-6 cycloalkyl)amino, or an amino acyl residue having structural formula

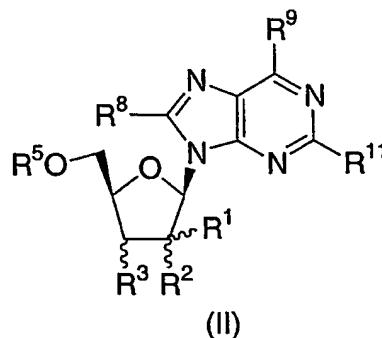


n is 0, 1, or 2;
 R11 is hydrogen, hydroxy, halogen, C1-4 alkoxy, amino, C1-4 alkylamino,
 10 di(C1-4 alkyl)amino, C3-6 cycloalkylamino, or di(C3-6 cycloalkylamino);
 R15, R16, and R17 are each independently hydrogen or C1-6 alkyl;
 R12 and R13 are each independently hydroxy, -OCH₂CH₂SC(=O)C1-4 alkyl,
 -OCH₂O(C=O)OC1-4 alkyl, -NHCHMeCO₂Me, -OCH(C1-4 alkyl)O(C=O)C1-4 alkyl,



R14 is hydrogen, C1-6 alkyl, C2-6 alkenyl, C2-6 alkynyl, C1-4 alkylamino, CF₃, or halogen; and
 R18 is hydrogen, C1-4 alkyl, or phenyl C0-2 alkyl.

20 2. The compound of Claim 1 of structural formula II:



or a pharmaceutically acceptable salt thereof;

wherein

- R1 is fluoromethyl or difluoromethyl;
- 5 R2 is hydroxy, fluoro, or C1-3 alkoxy;
- R3 is hydrogen, halogen, hydroxy, amino, or C1-3 alkoxy;
- R5 is hydrogen, P₃O₉H₄, P₂O₆H₃, or PO₃H₂;
- R8 is hydrogen, amino, or C1-4 alkylamino; and
- R9 and R10 are each independently hydrogen, halogen, hydroxy, amino, C1-4 alkylamino, di(C1-4 alkyl)amino, or C3-6 cycloalkylamino.

3. The compound of Claim 2 wherein

- R1 is fluoromethyl or difluoromethyl;
- R2 is hydroxy, fluoro, or methoxy;
- 15 R3 is hydrogen, fluoro, hydroxy, amino, or methoxy;
- R5 is hydrogen or P₃O₉H₄;
- R8 is hydrogen or amino; and
- R9 and R10 are each independently hydrogen, fluoro, hydroxy, or amino.

20 4. The compound of Claim 1 selected from the group consisting of:

- 6-amino-9-(2-C-fluoromethyl-β-D-ribofuranosyl)purine;
- 6-amino-9-(2-C-fluoromethyl-β-D-arabinofuranosyl)purine;
- 2-amino-9-(2-C-fluoromethyl-β-D-ribofuranosyl)-3,9-dihydropurin-6-one;
- 25 2-amino-9-(2-C-fluoromethyl-β-D-arabinofuranosyl)-3,9-dihydropurin-6-one;
- 2-amino-9-(2-C-fluoromethyl-β-D-ribofuranosyl)-3,9-dihydropurin-6-thione;
- 2,6-diamino-9-(2-C-fluoromethyl-β-D-ribofuranosyl)purine;

9-(2-C-fluoromethyl- β -D-ribofuranosyl)-6-methylaminopurine;
2'-C-(fluoromethyl)cytidine;
2'-C-(fluoromethyl)-5-methylcytidine;
2'-C-(fluoromethyl)uridine;
5 2'-C-(fluoromethyl)-5-methyluridine;
and the corresponding 5'-triphosphates;
or a pharmaceutically acceptable salt thereof.

5. The compound of Claim 4 which is
10 2-amino-9-(2-C-fluoromethyl- β -D-ribofuranosyl)-3,9-dihydropurin-6-one;
or a pharmaceutically acceptable salt thereof.

6. The compound of Claim 4 which is 6-amino-9-(2-C-fluoromethyl- β -D-ribofuranosyl)purine;
15 or a pharmaceutically acceptable salt thereof.

7. A pharmaceutical composition comprising a compound of
Claim 1 and a pharmaceutically acceptable carrier.

20 8. A method of treating RNA-dependent RNA virus infection
comprising administering to a mammal in need of such treatment a therapeutically
effective amount of a compound according to Claim 1.

9. The method of Claim 8 wherein said RNA-dependent RNA
25 virus infection is hepatitis C virus (HCV) infection.

10. The method of Claim 9 in combination with a therapeutically
effective amount of another agent active against HCV.

30 11. The method of Claim 10 wherein said agent active against
HCV is a 2'-C-Me-ribonucleoside; ribavirin; levovirin; thymosin alpha-1; interferon- β ;
an inhibitor of NS3 serine protease; an inhibitor of inosine monophosphate
dehydrogenase; interferon- α or pegylated interferon- α , alone or in combination with
ribavirin or levovirin.

12. The method of Claim 11 wherein said agent active against HCV is interferon- α or pegylated interferon- α , alone or in combination with ribavirin.

13. Use of a compound of Claim 1 for treatment of RNA-
5 dependent RNA virus infection in a mammal.

14. The use of Claim 13 wherein said RNA-dependent RNA virus infection is HCV infection.

10 15. Use of a compound of Claim 1 in the manufacture of a medicament for treatment of RNA-dependent RNA virus infection in a mammal.

16. The use of Claim 15 wherein said RNA-dependent RNA virus infection is HCV infection.